Supplementary data

Supplementary Figure Legends

Supplementary Fig. I. Akt phosphorylation was cytoprotective via upregulation of HIF-1α expression. (A) Densitometric analysis of Western blots in Fig. 1E showing increased phosphorylation of Akt (Ser-473) and Erk1/2 (Thr-202/Try-204), and Bcl-xl protein expression in ^{PC}MSCs with 2 cycles of 30-min I/R (Ť & Ŧ *vs* all other groups of cells $p<0.05$). (B) TUNEL staining showed higher number of apoptotic cells in the 30-min 3 cycles treated cell as compared with 30-min 1 cycle and 30-min 2 cycles treatment (C) Western blot showing elevated pAkt in ^{PC}MSCs which was abrogated by 50 μM Wort pretreatment of the cells. (D) IP-induced nuclear HIF-1α expression was abolished by Akt inhibition (Ť vs Ψ $p<0.01$; Ŧ vs Ψ p= *NS;* Ť vs Ŧ $p<0.01$). (E) Cytoprotective effect of IP was abolished by Akt inhibition as determined by LDH release assay (Ť & Ŧ *vs* all other groups of cells $p<0.05$). (F) RT-PCR showed upregulation of miR-210 in ^{PC}MSCs. Multiple cycles of I/R was more effective to induce miR-210 than single cycle of I/R.

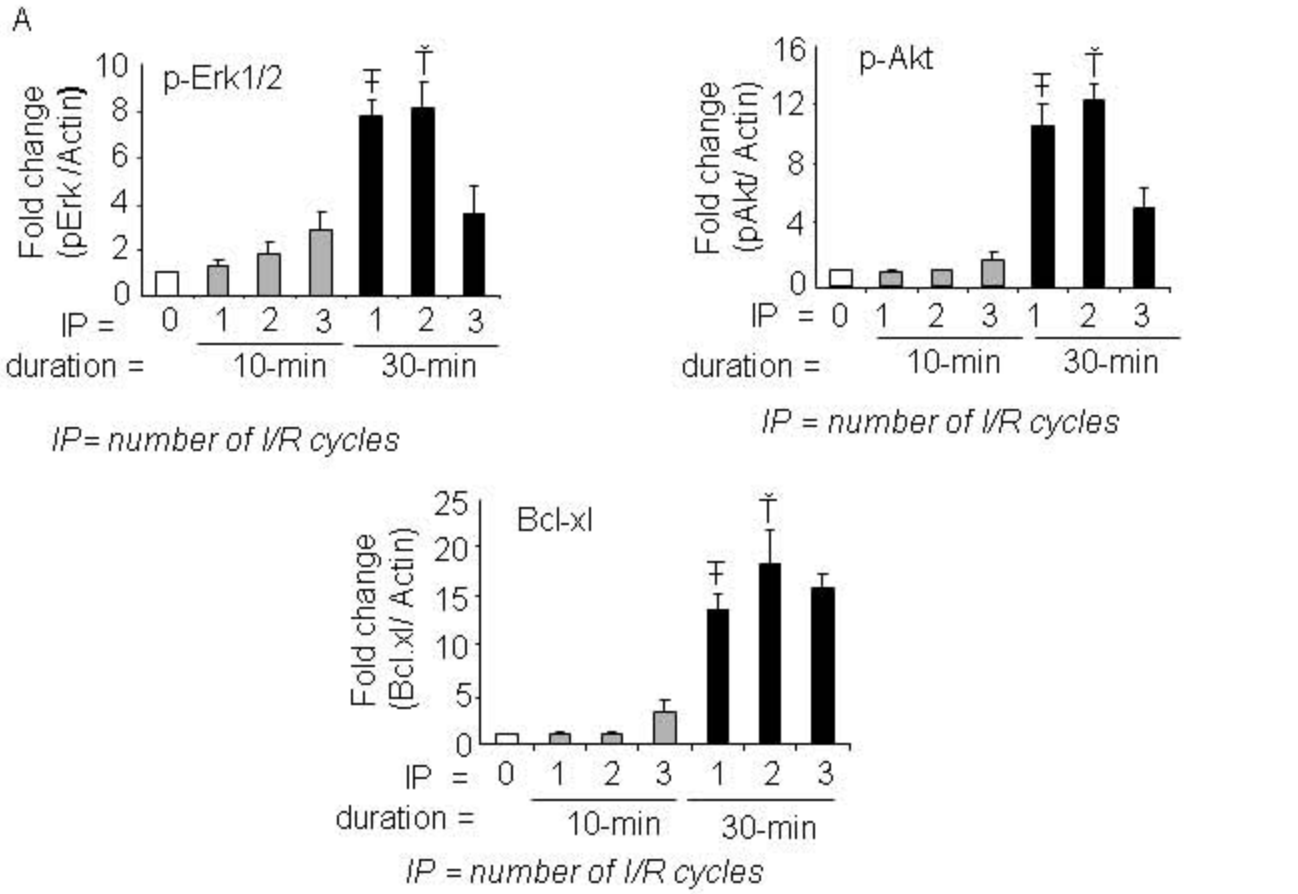
Supplementary Fig. II. IP with multiple cycles of brief I/R enhances miR-210 expression. (A) Knockdown of miR-210 increased apoptosis as examined by TUNEL staining (Ť & Ŧ *vs* all other groups of cells $p<0.05$) (original magnification x200, green= TUNEL$^+$ nuclei, blue= DAPI). (B) RT-PCR to show that pretreatment of the cell with 50 μM Wort inhibited IP-induced miR-210.

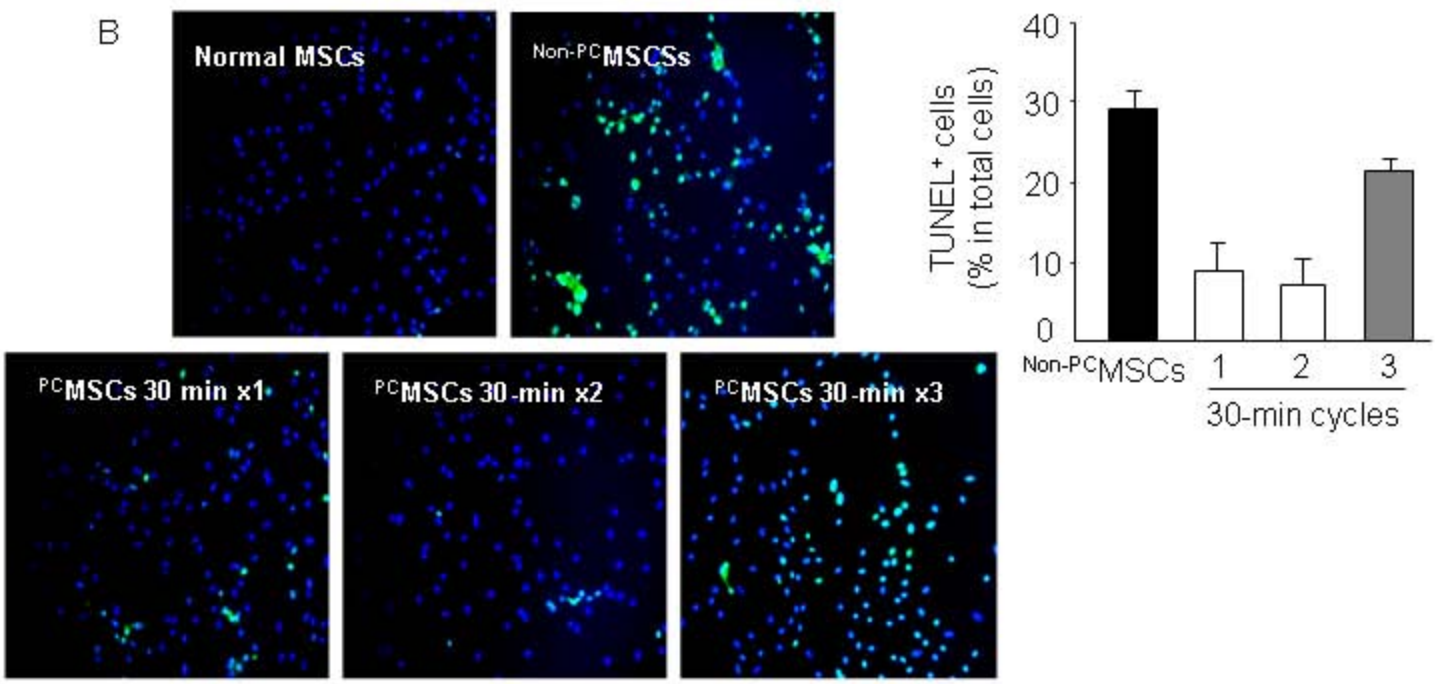
Supplementary Fig. III. The role of IP-induced HIF-1α in regulation of miR-210 expression and apoptosis. Knockdown of HIF-1α and abrogation of miRNAs lead to higher TUNEL positivity in ^{PC}MSCs transfected with HIF-1 specific siRNA (magnification x200, green= TUNEL$^+$ nuclei and blue= DAPI; Ť vs Ŧ $p<0.01$).

Supplementary Fig. IV. (A) Real-time PCR showing higher level expression of *Casp8ap2* in miR-210 abrogated ^{PC}MSCs upon exposure to 6h anoxia ($^*p<0.01$ vs controls). (B) Fluorescence images showing TUNEL positive cells in ^{PC}MSCs and $^{non\text{-}PC}$MSCs transfected with *Casp8ap2* siRNA as compared with their counterparts with Sc siRNA. Quantitative data is shown in Fig. 3F.

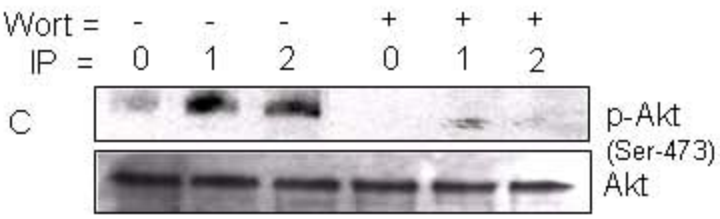
Supplementary Table-I: List of the antibodies used.

Antibody	Dilution used	Source
Anti-p-Akt (ser-473)	1:500	Cell Signaling Technology
Anti p-p44/p42 (Thr202/Tyr204)	1:500	Cell Signaling Technology
Bcl-xl	1:1000	Cell Signaling Technology
Anti HIF-1α	1:1000	Novus Biological
Anti actin	1:2000	Santa Cruz Biotechnology

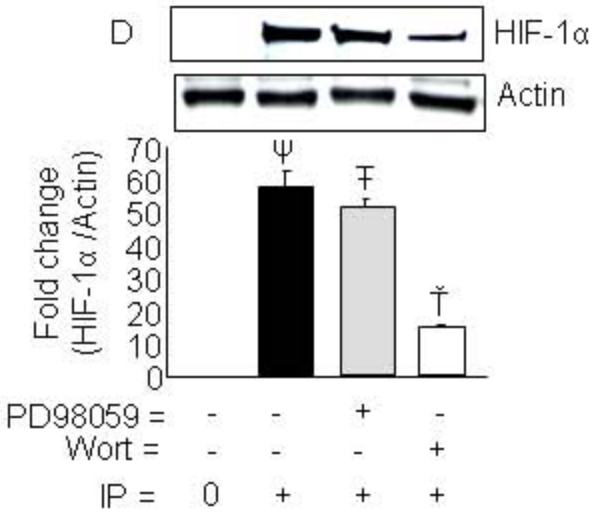




Supplementary Figure I (con)



IP=number of 30 min I/R cycles



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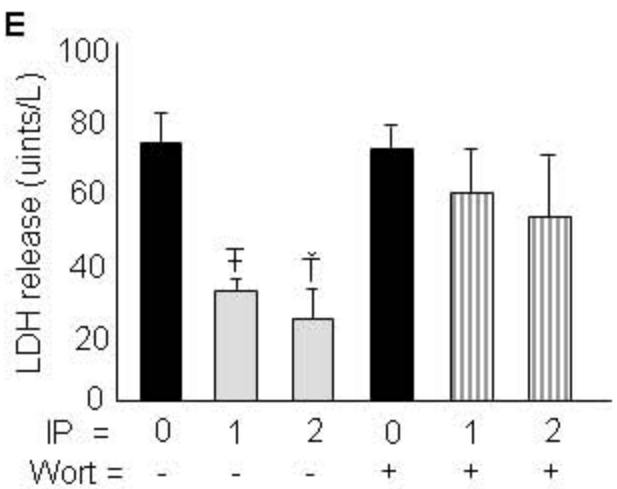
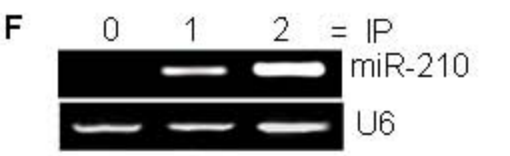
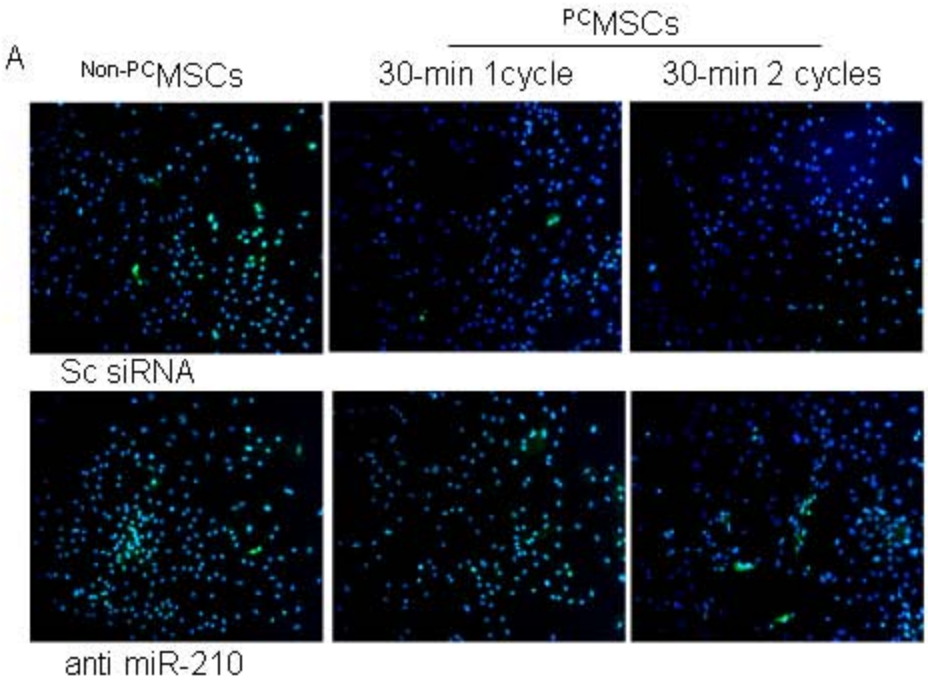
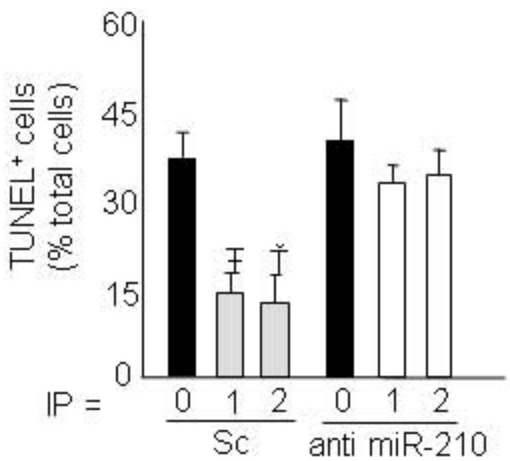


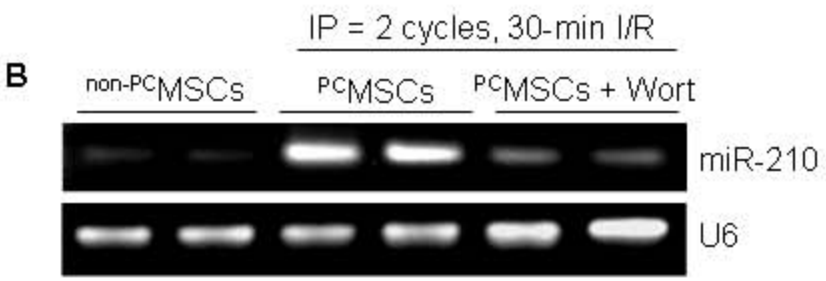
IP=number of 30-min I/R cycles



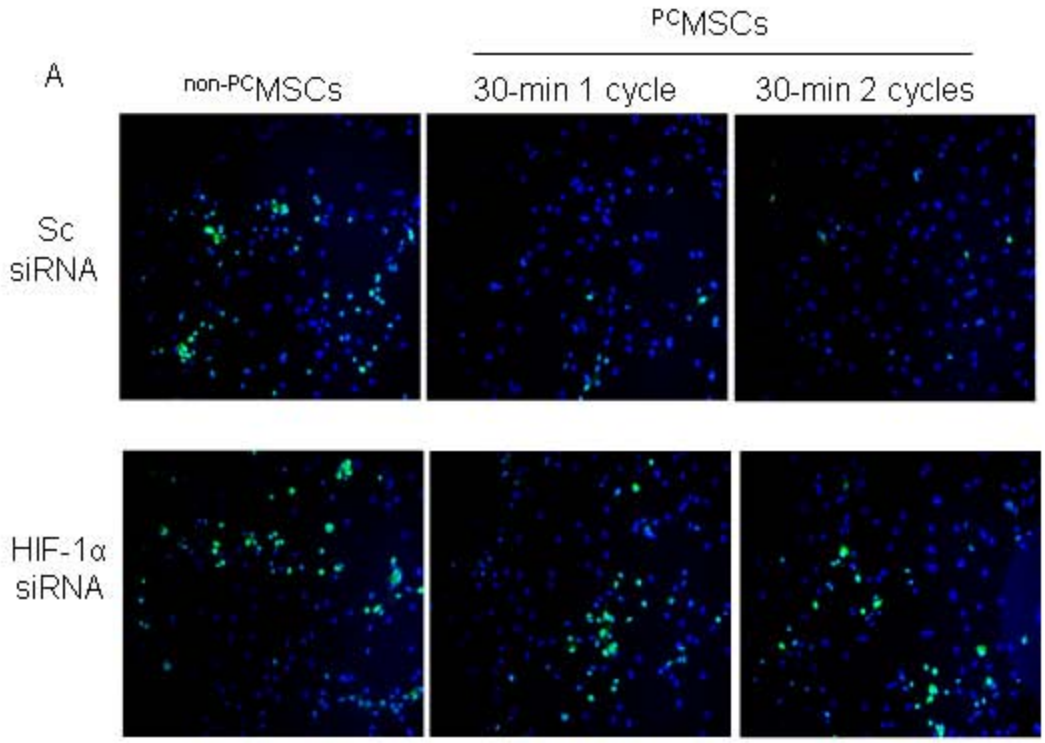
IP = number of 30-min I/R cycles

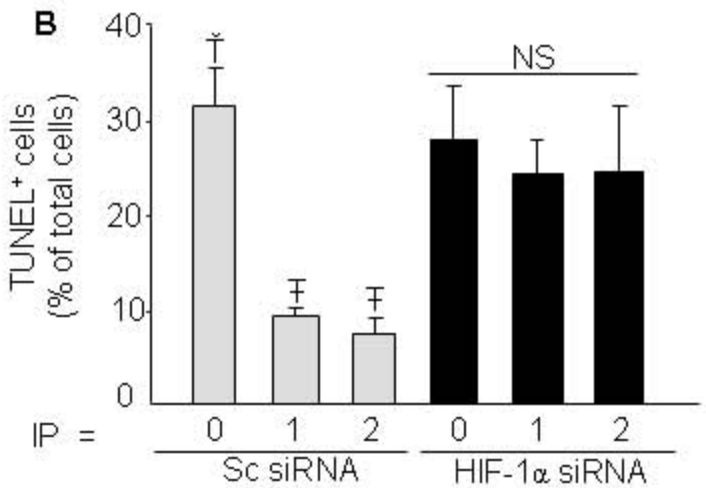

A
PCMSCs
Non-PCMSCs
30-min 1cycle
30-min 2 cycles
Sc siRNA
anti miR-210


TUNEL+ cells
(% total cells)
60
45
30
15
0
IP =
0 1 2
0 1 2
Sc
anti miR-210
IP (number of 30-min I/R cycles)

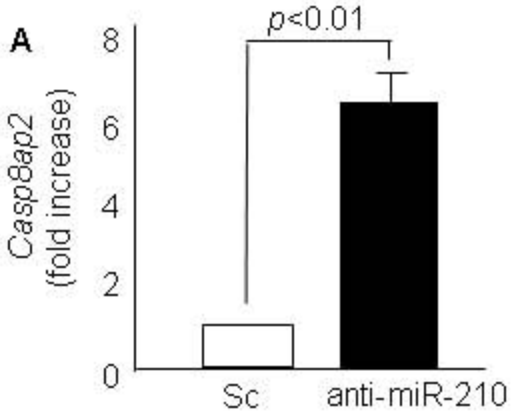

B
IP = 2 cycles, 30-min I/R
non-PCMSCs
PCMSCs
PCMSCs + Wort
miR-210
U6

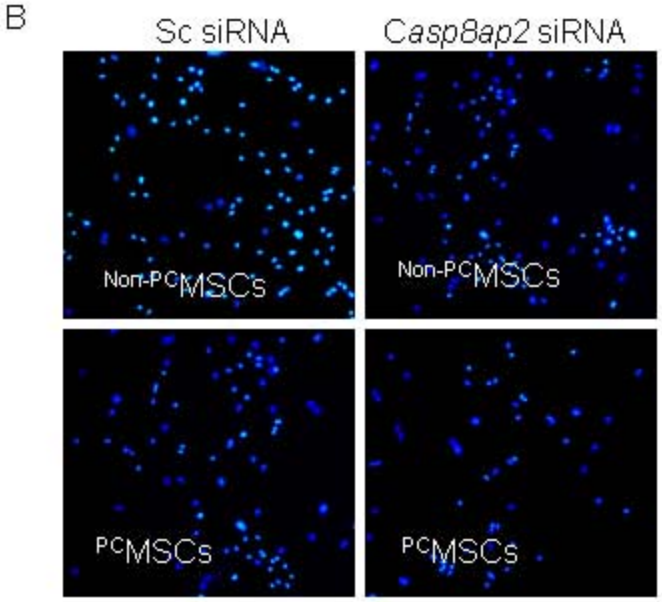
Supplementary Fig. III Kim et al. 2009





IP= number of 30-min I/R cycles


A
p<0.01
Casp8ap2
(fold increase)
8
6
4
2
0
Sc
anti-miR-210


B
Sc siRNA
Casp8ap2 siRNA
Non-PCMSCs
Non-PCMSCs
PCMSCs
PCMSCs